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α 2-dystrobrevin siRNA (m): sc-43324

BACKGROUND

Dystrobrevins are protein components of the dystrophin complex, whose disruption leads to Duchenne muscular dystrophy and related diseases. α -dystrobrevin is a dystrophin-related and -associated protein that is involved in synapse maturation and is required for normal muscle function. α -Dystrobrevin is a component of the dystrophin glycoprotein complex. It is localized to the cytoplasmic side of the sarcolemma and is highly concentrated at the neuromuscular junctions in skeletal muscle. The insertion of 57 amino acids by alternative splicing accounts for the increase in molecular mass of α -Dystrobrevin 1 in skeletal and cardiac muscle compared with brain and lung. α -Dystrobrevin containing complexes are found in endothelial and smooth muscle cells, while β -Dystrobrevin containing complexes are present at the basal region of renal epithelial cells. Additionally, β -Dystrobrevin is found in neurons and is highly enriched in postsynaptic densities. Alternative splicing of α -Dystrobrevin produces γ -Dystrobrevin (isoform 5), δ -Dystrobrevin (isoform 7), ϵ -Dystrobrevin (isoform 6) and ζ -Dystrobrevin (isoform 8). Additional isoforms may also exist.

REFERENCES

1. Blake, D.J., et al. 1998. β -Dystrobrevin, a member of the dystrophin-related protein family. *Proc. Natl. Acad. Sci. USA* 95: 241-246.
2. Blake, D.J., et al. 1999. Different dystrophin-like complexes are expressed in neurons and glia. *J. Cell Biol.* 147: 645-658.
3. Loh, N.Y., et al. 2000. Assembly of multiple Dystrobrevin-containing complexes in the kidney. *J. Cell Sci.* 113: 2715-2724.
4. Gieseler, K., et al. 2001. Molecular, Genetic and physiological characterization of dystrobrevin-like (dyb-1) mutants of *Caenorhabditis elegans*. *J. Mol. Biol.* 307: 107-117.
5. Newey, S.E., et al. 2001. A novel mechanism for modulating synaptic gene expression: differential localization of α -Dystrobrevin transcripts in skeletal muscle. *Mol. Cell. Neurosci.* 17: 127-140.
6. Enigk, R.E., et al. 2001. Cellular and molecular properties of α -Dystrobrevin in skeletal muscle. *Front. Biosci.* 6: 53-64.

CHROMOSOMAL LOCATION

Genetic locus: Dtna (mouse) mapping to 18 A2.

PRODUCT

α 2-dystrobrevin siRNA (m) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see α 2-dystrobrevin shRNA Plasmid (m): sc-43324-SH and α 2-dystrobrevin shRNA (m) Lentiviral Particles: sc-43324-V as alternate gene silencing products.

For independent verification of α 2-dystrobrevin (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-43324A, sc-43324B and sc-43324C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNases and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

α 2-dystrobrevin siRNA (m) is recommended for the inhibition of α 2-dystrobrevin expression in mouse cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μ M in 66 μ l. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

α -Dystrobrevin (D-9): sc-271630 is recommended as a control antibody for monitoring of α 2-dystrobrevin gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz[®] Mounting Medium: sc-24941 or UltraCruz[®] Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor α 2-dystrobrevin gene expression knockdown using RT-PCR Primer: α 2-dystrobrevin (m)-PR: sc-43324-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.